EPIDEMIOLOGICAL STUDY ON CAMEL MASTITIS IN NORTH KORDOFAN STATE, SUDAN

By

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DEDICATION

TO:

my

parents,

brothers,

and

sisters.

PREFACE

This work was carried out in the Department of Preventive Medicine and Veterinary Public Health, Faculty of Veterinary Medicine, University of Khartoum, under the supervision of Dr. Tawfig ElTigani Mohamed. The material presented is original and has not been submitted to any University.

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ABSTRACT

Mastitits had economic importance which resulted in reduced milk production. The diagnosis of clinical mastitis either by visual examination of milk and udder for apparent signs of mastitis or culturing and screening tests for sub-clinical mastitis (Quinn *et al*., ١٩٩٤).

This study was carried out in North Kordofan State to determine the prevalence of mastitis in lactating she-camels on bases of the questionnaire survey and bacterial isolates. Also to evaluate the W.S.T and S.C.C. in diagnosis of mastitis and to determine the susceptibility of various bacterial isolates to antibiotics which were used for mastitis treatment.

A total of ١١٨ camel owners were interviewed and the general investigation showed ٩٩٪ of owners confirm that mastitis was present in lactating she-camels, ٩٧٪ and ٩٢,٣٧٪ did not clean the udder or their hands before milking, respectively. Also ٨٥,٠٩٪ of them did not receive Veterinary and extension services, ٩٦,٦١٪ used the anti-suckling devices and ٨٥,٠٥٪ showed that presence of anti-sucking devices without milking of lactating she-camel as the main causes of mastitis. Moreover, sampling questionnaire revealed that ١٠٠٪ of lactating she-camels were infested with ticks in their udder and ٥٦,٦٧٪ had teats lesions.

Out of ٦٠ lactating she-camels, ٢١٦ quarter milk samples were collected and examined by W.S.T., S.C.C. and bacteriological examinations. The results of positive tests were ١٦,٥%, ١٨,٦٦٪ and ٤٥,٨٣٪ respectively. furthermore, the prevalence of sub-clinical mastitis represented ٤٥,٣٧٪ and clinical mastitis ٠,٩٣٪. In this study the bacterial isolates were *Staphylococcus* (٨٠,٣٠٪), *Streptococcus* (١,٥٢), *Corynebacteria* (٣,٠٣٪), *Bacillus* (٩,٠٩٪) and *Pasteurella* (٦,٠٦٪). Furthermore, the species of *Staphylococcus* isolated were *Staphylococcus aureus* (٢٢,٧٥٪), *S. hyicus* (٣,٠٣٪), *S. intermediatius* (٧,٥٦٪). *S. delphini* (٣,٠٣٪) *S. epidermidus* (١٢,١٢٪), *S. simulans* (٦,٠٦٪),S. *kloosii* (٤,٥٥٪), *S. chromogenes* (١,٥٢٪), *S. lentus* (٣,٠٣٪), *S. lugdunensis* (٣,٠٣٪), *S. sacchrolyticus* (١,٥٢٪), *S. saprophyticus* (٣,٠٣٪), *S. haemolyticus* (٦,٠٦٪) and *S. carnosus* (١,٥٢٪).

This study confirmed the strong relationships between S.C.C. and W.S.T. with bacteriological examinations. The antibiotic sensitivity test revealed the high sensitivity of the isolates to *Ciprofloxacin, Gentamycin*, *Offlaxacin Cephalexine*, *Tetracyline* and *Co-trimoxazole* and intermediate sensitivity to *Ampicillin* and *Cefotaxime*. However, the tested organisms confirmed the resistance to *Coloxacillin* and *Lincomycin.*

الخلاصه

التهاب الضرع له اهميه اقتصاديه تتمثل في انخفاض انتاجية اللبن . ويـتم تـشخيص التهـاب الضرع السريري بالفحص بالعين المجرده لملاحظة التغيرات في اللبن والضرع او بالاختبارات المسحيه الي جانب العزل البكتيري لالتهاب الضرع تحت السريري.

أجريت هذه الدراسه بولاية شمال كردفان وذلك لمعرفة مدي انتشار التهاب الضرع في النـوق الحلوبـه وذلك اعتماداً علي التقصي عن المرض، والعوامل المهيئه علي حدوثه، والعزل البكتيري، وايضاً تقيـيم مدي فعالية اختبار التعداد الكلي للخلايا الحيويه واختبار الويت سايد فـي تـشخيص التهـاب الـضرع. ولتحديد مدي حساسية البكتريا المعزوله للمضادات الحيويه المستخدمه في علاج التهاب الضرع.

قد تم مقابلة ١١٨ صاحب ابل وكانت اراءهم عن التهاب الضرع كالاتي ٩٩% اكدوا علي وجود التهاب الضرع في النوق الحلوبه و٤٦،٩٧% من الحلابين لم ينظفوا الضرع قبل عملية الحليب و ٩٢،٣٧% لم يغسلوا اياديهم قبل عملية الحليب. ايضاً ٨٥,٠٩% من رعاة الابل لم يتلقوا خدمات بيطريه وارشاديه ، و ٩٦،٦١% بيستخدموا الجهاز المانع للرضاعه و ٨٥،٠٥% برهنوا ان وجود الجهاز المانع للرضـاعه

مع عدم عملية الحليب من اهم مسببات التهاب الضرع. اضافة الي ذلك اوضح استبيان جمع العينات ان ١٠٠% من النوق الحلوبه بها قراد في الضرع ٥٦،٩٧% بها افات في حلمات الضرع.

ان من مجموع ٦٠ ناقه حلوب تم اخذ ٢١٦ عينة لبن علي حسب ارباع الضرع وفحصت باختبار الويت سايد ،اختبار التعداد الكلي للخلايا الحيويه والاختبارات البكتريولوجيه. فكانت النتائج الموجبه للاختبارات السابقة علي التوالي (١٦،٥%)، (١٨،٦٦%)، (٤٥،٨٣%) اضافه الي ذلك يمثل معدل انتشار التهـاب الضرع تحت السريري (٤٥،٣٧%) والسريري (٠،٩٣%)

في هذه الدراسه البكتريا التي تم عزلها تتضمن الاتي: المكـورات العنقوديـه (٨٠،٣%) والمكـورات السبحية ١،٥٢% والوتديات ٣،٠٣% ، العصويات الباسيلية ٩,٠٩% والباستيوريا ٦،٠٦%. اضافه الي ذلك، ان انواع المكورات العنقودية التي تم عزلها تتمثل فـي التـالي: المكـورات العنقوديـة الذهبيـه (٢٢،٧٥%)، المكورات العنقودية الخنزيريه (٣،٠٣%)، المكـورات العنقوديـة البـشرويه ١٢،١٢%، المكورات العنقودية المتشابهه ٦،٠٦%، المكورات العنقودية الكلوسية ٤،٥٥%، المكـورات العنقوديـة الصبغية ١،٥٢%، المكورات العنقودية العدسية ٣،٠٣%، المكـورات العنقوديـة الـسنجابيه ٣،٠٣%، المكورات العنقوديه السكريه ١,٥٢%، المكورات العنقودية الرميه ٣،٠٣%، المكورات العنقوديه حالـة الدم ٦،٠٦% والمكورات العنقودية اللحمية ١،٥٢%.

اكدت هذه الدراسه علاقات قوية بين كل من اختبار التعداد الكلي للخلايا الحيويه واختبار الوايت سايد مع الاختبارات البكتريولوجية.

اوضح اختبار الحساسية للبكتريا المعزولة حساسية عاليه لكـل مـن سبروفلوكـساسين، جنتاميـسين ، اوفلاكساسين، سيفلاكسين، تتراساكلين، كوا-تراموكسازول، وحساسية متوسطة للامبسلين والسيفوتاكسيم. اضافة الي ذلك اظهرت البكتريا المعزوله اقل حساسية لكل من الكولوكساسلين و اللينكوماسين.

INTRODUCTION

In the family *camelidae* there are two genera *lama* and *camelus*. The second genus is classified to *camelus dromedaries* (one humped camel) and *camelus bactrianus* (two humped camel). The total population of camel in the world is about ١٧,٤٧ million, the Sudan has ٢,٥٧ million which is about ١٤,٧٪ (Animal Resources National Statistic, ١٩٨٩). Advance estimate was recorded by FAO, (١٩٨٩) as ٣,١ million. However, the one humped camel in the world is estimated as ١٧ millions heads, ١٥ millions of it are found in the Arab countries. (Haroun,١٩٩٠). Sudan and Somalia constitute about ٥٠٪ of all camels in the world and ٥٥٪ of all *dromedaries*. Also, Sudan holds ٣٢٪ of camels of Africa and ٢٠٪ of the world camel population. (Wilson, ١٩٨٤, FAO report, ١٩٧٧). While the two humped camel is about ١,٧ million heads in the natural habitat in Asia (Fassi-Fahri, ١٩٨٧).

Camels are distributed in the Sudan north of ١٤ N° latitude in the Western and ١٦N° latitude in the Eastern region (Mohamed, ١٩٩٢). The system of camel raising and husbandry in the Sudan is a traditional nomadic one. Camels occupy the desert and semi-desert of Northern Kordofan and Darfur, Butana area and Kassala to the Red sea Hills. (Mohamed, ١٩٩٢, Yagoub, ٢٠٠٣). The camels in Kordofan are raised by their expert owners of Kababeish, Shenabla, Hamar, Kawahla and Dar Hamid tribes. Their camels migrate to the South of Kordofan

during the dry season. After the beginning of the rainy season “Rushash” all camels migrate to the North of Kordofan avoiding the harmful effect of biting files and muddy conditions in South Kordofan. Camels of Northern Darfur inhabit and environment similar to that of Kordofan, they have also similar seasonal migration, however the most important camel owing tribes of Northern Darfur are the medoub, zagawa, zayadia and other minority tribes. In Eastern Sudan most camel owing tribes are Hadendawa, Rashida and Beni Amir tribes. The types of camels in the Eastern region are Bishari and Anafi. In Butana area famous tribes owning camels are Shukria, Lahwien and Ruffaa. (Yagoub, ٢٠٠٣). Camels in Sudan classified to the riding (light) such as, Bishari and Anafi, or pack (heavy) camel, such as, Arab and Rashaida camel according to the function which being performed (Elamin, ١٩٧٩).

The FAO, (١٩٩٢), report indicated that the food supply must be improved, both in quantity and quality. Therefore, it is well known that camels play major roles in improving the socio-economic status and survival of desert dwellers and are major sources of protein and energy for them. This is due to a number of specific anatomical and physiological characteristics of the camel as well as low susceptibility to diseases (Schwartz and Dioli, ١٩٩٢).

However, during the last decades the diseases has been reported from a number of camels in the Sudan (Abdurhman *et al.,* ١٩٩٥; Obied *et al*., ١٩٩٦). On the other hand, udder infection was considered as one of the main constraints for camel rearing. For instance, it has been noticed in the slaughter house, that early culling

of female in Iraq is attributed to chronic mastitis and infertility (Al-Ani and Shareefi, ١٩٩٧).

According to Abdel Gadir, (٢٠٠١). The Somatic Cells Counts (SCC) of normal and mastitic camel milk requires more investigations. Also , an attempt should be made to increase awareness of camel owner on the importance and impact of udder infection on public health and milk yield. Therefore, the objectives of this study are :

- To determine the prevalence of mastitis in lactating she-camel on bases of the questionnaire survey and bacterial isolates.
- To evaluate White side test (WST) and Somatic cell counts (SCC) based on microbiological examinations for the detection of camel mastitis.
- To analyze the risk factors that affects the occurrence of camel mastitis.
- To determine the sensitivity of various bacterial isolates to antibiotics which were used for mastitis treatment.

CHAPTER ONE

LITERATURE REVIEW

١,١ Economic importance of the camel

Camels products are milk, meat and by products such as, wools and hides. The camel milk in Kordofan, is considered the main source of food and water for Kababesh tribes during the winter migration to the far north "Guzzu" grazing in the desert. However, camels are used for riding, racing, packing and pulling water from deep wells (Mohamed *et al,* ١٩٩٠, Yagoub, ٢٠٠٣). Camel milk contains the necessary proteins, sugars, fats, minerals and vitamins for the calves and is a valuable food for people, also camel milk is a rich source of vitamin C for the desert people who are unable to get it from other sources. (Schwartz and Dioli, ١٩٩٢, Wilson, ١٩٩٨).

Nomadic camel owners of the Sudan are utilizing wool and hair for making their temporary lodgings. Bags for storing drinking water and fermented souring milk are made from camel hides. Beds, mats to rest on and bags to store and the house decoration are made from camel hides and hair. Socially, many camels are a sign of wealth and importance in the nomadic rules (Yagoub, ٢٠٠٣).

Camels constitute ٦ % of the livestock producing meat and milk in Sudan. about ٨ million heads of animal were slaughtered annually for meat and camels meat constitute ٨,٨٪ of that group (Haroun, ١٩٩٠, Majed, ٢٠٠٠). Sudan is capable of producing more than ١٠٠٠٠ tons of camels meat annually (AOAD,١٩٨٣). Camels contribute in the gross national product of Sudan through exportation to the Arab

countries of meat and racing camels as shown by governmental documents exported livestock adds more than ٨٠ million U.S dollars of the national economy of which camels constitute ٢٥٪ (Haroun, ١٩٩٠, Ahmed Gotbi, ١٩٨٩).

١,٢ Diseases of camel

١,٢,١ Viral diseases

In North Kordufan State, an out break of rabies occurred in (١٩٩٨) among camels near Khor Taggat, total number of animals affected was ٥٢ camels, ١١ of them died (Annual reports, Elobied Veterinary Research laboratory records, ١٩٩٨). Khalaffalla (١٩٩٨) reported an out break of camel pox in Butana area with mortality rate of ١-٢٪ in camel calves. Camel contagious ecthyma was also reported by the same author with mortality rate of ٨,٨٪. Antibodies to blue tongue virus were detected by Abu Elzen (١٩٨٥) with prevalence rate of ١٦,٦٪.

١,٢,٢ Parasitic diseases

Abu Samra (١٩٨٦) studied the incidence of sarcoptic mange in ٣٣،٠٠٠ camels from Kassala, Red sea, Nile and Northern provinces. ٥٥,١٪ of these camels were found infected. Musa and Osman (١٩٩٠) reported an out break of a suspected tick paralysis in Sudanese camels. Musa *et al* (١٩٨٩) reported camel myasis caused by *Cephalopina titilator* in ٤٤ cases from Nyala slaughter house. The larvae migrate in the nasal sinuses causing severe irritation. Haemonchosis is known by owners as "Hulla". Addel Gaffar *et al* (١٩٨٤) investigated camel haemonchosis in Eastern Sudan and reported ٦٧٪ infection rate. Mahmoud and Gray (١٩٨٠) stated that the

major protozoan disease affecting camels is trypanosomiais caused by infection with trypanosoma evansi.

١,٢,٣ Fungal disease

In Sudan Fadle Elmola, *et al* (١٩٩٤) isolated *Trichophyton verrucosum* as the causative agent of disease in camel .

١,٢,٤ Bacterial disease

Anthrax disease was reported from Darfur in both man and camels (Musa *et al*, ١٩٩٣, Yagoub *et al.,* ١٩٩٠). Detected antibodies to Brucella abortus from the Eastern Region of the Sudan as ٦,٩٥٪ in males and ١٣,٧٦٪ in females. Camel-calf diarrhea was investigated by many researchers. Abass (١٩٩٣) reported that camel ageing ٧ days to ٥ months were affected , with mortality rate of ٢٣٪ it attributed the cause to *Salmonella* infection.

١,٣ Definition of mastitis

Mastitis refers to inflammation of the mammary gland and can be caused by physical or chemical agents but the majority of causes are infectious agents usually caused by bacteria. (Quinn *et al.*, ١٩٩٤).

١,٤ Types of mastitis

There are two types of mastitis, clinical and sub-clinical mastitis.

١,٤,١ Clinical mastitis

Includes peracute, acute and chronic mastitis.

١,٤,١,١ Peracute mastitis

Characterized by swelling, pain, heat and abnormal secretions in the mammary gland and accompanied by signs of systemic disturbance such as fever, depression and anorexia or septicemia.

١,٤,١,٢ Acute mastitis

Changes in the mammary gland are similar to those of peracute mastitis but the systemic signs are less severe.

١,٤,١,٣ Chronic mastitis

There are no systemic signs and very few external signs of change in the udder, but abnormal secretion from the gland occurs intermittently.

١,٤,٢ Sub-clinical mastitis

The infection in the mammary gland is detectable only be bacterial culture or by tests to demonstrate a high leukocyte count in the milk, such as S.C.C. or C.M.T. test. There is obvious change in the appearance of the milk (Quinn *et al*., ١٩٩٤).

١,٥ Pathogens causing camel mastitis

Saad and Thabet (١٩٩٣) collected milk samples randomly from clinically healthy she-camel and examined it for bacteriological quality and mastitis. The test revealed the presence of *Staphylococcus aureus*, *Micrococcus spp*. Coliform organisms, *Bacillus cereus* and *Pseudomonas aeruginosa. Streptococcus agalactiae* and *Staphylococcus aureus* were the main causes of camels mastitis (Obied *et al*., ١٩٩٦, Al-Ani and Alshareefi, ١٩٩٧).

E .coli has been reported as major pathogen causing mastitis in camel. Further more, *E.coli, Pseudomonas aeruginosa, Klebsiella pneumoniae* have been isolated from both clinical and sub-clinical camel mastitis. (Saad *et al.,* ١٩٩٣; Abdurahman *et al*. ١٩٩٥; Bekele and Molla, ٢٠٠١; Kapur *et al.,* ١٩٨٢; Quandil and Oudar, ١٩٨٤; and Ejakee, ١٩٩٨). On the other hand, *Proteus vuLvaris* and *Aerobacter spp*. have been isolated from normal camel udder tissue and milk, respectively (Obied *et al* ., ١٩٩٦; and Ejakee, ١٩٩٨).

In Saudi Arabia, Ramadan *et al*, (١٩٨٧) described unilateral chronic mastitis by *Staph. aureus* and *Pasteurela haemolytica* in three lactating camels due to obstruction of the teat canals by keratin. In India Kapur *et al*, (١٩٨٢) recorded a case of peracute mastitis caused by *Klebsiella spp*. and *E.coli* that resulted from contamination during udder surgery.

Relationship between occurrence of camel mastitis and presence of *Corynebacterium spp.,* and *Bacillus spp*. were recorded in Iraq (Al-Ani and Alshareefi, ١٩٩٧), Egypt (El. Jakee, ١٩٩٨), and Ethiopiupia (Almaw and Molla, ٢٠٠٠; Bekele and Molla, ٢٠٠١).

Nuha, (٢٠٠١), isolated *Staphylococcus chromgenes, S. lugdunensis* and *S. saccharolyticus* from camel milk. Amel, (٢٠٠٣) and Suheir (٢٠٠٤) isolated *Staphylococcus saprophyticus, S. carnosus, S. Kloosii, S. delphini* and *S. lentus* from camel milk. Also, Elayis,(٢٠٠٤), isolated *S. haemolyticus* from mastitic bovine milk .

١،٦ Risk factor of camel mastitis

The heavy tick infestation of the udders, the use of anti-suckling devices and treatment of infected quarter by cauterization have been considered as predisposing factors for camel mastitis (Abdurahman *et al*., ١٩٩٥; Obied *et al*., ١٩٩٦).

١،٧ Epidemiology of mastitis

Mastitis in the camel has been reported from different countries, such as Somilia (Arush *et al.,* ١٩٨٤; Abdurahman *et al.,* ١٩٩١); Egypt (Moustafa *et al*., ١٩٨٧; Karmy, ١٩٩٠) Saudi-Arabia (Barbour *et al.,* ١٩٩٥; Hafez *et al .,* ١٩٨٧; Ramadan *et al.,* ١٩٩١); United Arab Emirates (Quandil and Ouadar, ١٩٨٤); Ethipiupia (AlMaw and Molla; ٢٠٠٠; Salah,٢٠٠٠; Bekele and Molla; ٢٠٠١), and Kenya (Younan *et al*., ٢٠٠١).

In Sudan, camel mastitis was reported from different parts of Sudan. for instance, the first report of camel mastitis was by Obied (١٩٨٣). Then followed by (Bakhiet, Agab and Mamoun, ١٩٩٢. Agab, ١٩٩٣., Salwa, ١٩٩٥. Abdurhman *et al*., ١٩٩٥).

Agab and Abbas (١٩٩٨) reported that the percentage of mastitis was ٤-٢١٪ in total of ٣٧٣١ she-camels examined in eastern Sudan. Moreover, camel mastitis was also report as ١١٪ of all animal diseases diagnosed at Atbra Veterinary Hospital during the period from July ١٩٩١, to June ١٩٩٣.(El Ghali and El Hussein, ١٩٩٥).

Obeid *et al.,* (١٩٩٦), identified clinical mastitis in camel which was characterized by hardening and swelling of udder, pain in palpation and visible alteration of colour and consistency of milk, thus clinical mastitis can be detected by

examination of udder and/or of the milk. Also sub-clinical mastitis was reported in Sudan, indicated that the term sub-clinical mastitis in camels refers to the existence of inflammation in the absence of gross signs and can be detected by indirect tests. Such as, California mastitis test (CMT) and somatic cell count (SCC) as well as Microbiological examinations. (Quinn *et al*., ١٩٩٤).

١,٨ Diagnosis of camel mastitis

Mastitis should be regarded as a herd problem and the methods of investigation and diagnosis should reflect this fact. The diagnostic methods include: total and leukocyte cell counts for both herds and individual she-camel milk, direct chemical tests, microbiological investigation to determine the major pathogens causing mastitis in a herd and the percentage of sub-clinical carrier. Obied *et al*, (١٩٩٦) determined the leukocyte count of she-camel milk and found that, neither a significant correlation between the leukocyte count of milk and the isolated bacterial species nor a significant variation of leukocyte content during different stages of lactation. The same authors found strong correlation between leukocyte count and the C.M.T. scores. Samples showing count below ٥٠٠،٠٠٠ cell/ml were considered negative while those above ٥٠٠،٠٠٠ cell/ml were positive. Bagadi (١٩٦٦) stated that samples which showed an increased number of cells(٥٠٠،٠٠٠ or more/ml) with or without isolation of organism were regarded as suspicious to mastitis and those with neither cells or organisms were discarded as negative samples. The increase of leukocyte count in almost all cases of mastitis indicated tissue injury which was followed by changes in the milk (Radostits, Blood and

Gay, ٢٠٠٠). Saad and Thabet (١٩٩٣) have reported a strong correlation between white side test (WST) and bacteriological results of camel milk samples. American Public Health association (١٩٦٠), reported that variations of the White side test include the Colifornia Mastitis Test (CMT) in which a solution of surface active agent is substituted for the alkali of the original method. Clinical mastitis can be detected by examination of udder and /or of the milk also using indirect tests. Bacteriological examinations of the milk, Cloifornia mastitis test (CMT) and Somatic cell count (SCC) have been used as diagnostic tools to detect sub-clinical mastitis in camels (Quinn *et al*., ١٩٩٤).

١,٩ Treatment of camel mastitis

The antibiotic susceptibility patterns of the major pathogens (*Staphylococcus aureus, Pasteurela haemolytica, E .coli, Micrococcus spp, Bacillus cereus, Klebsiella pneumoniae* and *Streptococcus agalactiae*), so that effective treatment can be administered (Quinn, ١٩٩٤).Buxton and Fraiser (١٩٧٧) reported the resistance of *Staphylococcus aureus* to Penicillin and Tetracycline. Mc.Donald and Anderson (١٩٨١), reported the sensitivity of *S. aureus* to *Cephaloxin, Clozacillin, Gentamycin* and *Ampcillin*. Where as Gentilini *et al.*, (٢٠٠٢) reported sensitivity of Coagulase negative Staphylococci to Gentamycin and Ampicillin. Abdelgadir (٢٠٠١), reported that *Staphylococcus aureus* from camel's milk was susceptible to Tetracycline, Gentamycin and highly sensitive to Ampicillin. Also he found that *S. hyicus and S. intermedius* were highly sensitive to Tetracycline and Gentamycin.

CHAPTER TWO

MATERIALS AND METHODS

٢,١. Sterilization

٢,١,١ Sterilization of Equipments

Petridishes, test tubes, flasks and pipettes were sterilized in the hot air oven at ١٦٠°C for one hour. Glass ware such as Bijou, universal and MC. Cartney bottles were sterilized in the autoclave at ١٥ pounds for ١٥ minutes at ١٢١°C. Instruments such as scissors, forceps and scalpels were sterilized in the hot air oven at ١٦٠°C for one hour. (Merchant and Packer, ١٩٧١).

٢,١,٢ Sterilization of culture media and solutions

The culture media and saline solution were sterilized in the autoclave at ١٥ pounds for ١٥ minutes ١٢١°C (Barrows and Feltham, ١٩٩٣).

٢,٢ Area of study

٢,٢,١ Description of area of study

Surveys for camels mastitis were carried out in Kordofan State (North part of Kordofan) that lies between latitudes ٩ -١٦,٨°N and ٢٧-٣٢°E. The area is sandy with ridges " Gaezau". Some vallies, seasonal water pools and clay plains are scattered in the state. However, in the North Kordofan the area being desert and semi-desert, the camels owners live a nomadic life, migrating from place to another searching for water and grass.

٢,٢,٢ Vegetations

The vegetation of the area is composed of trees and grasses of different densities. The main species of trees is *Leptadenia ahaeterophylla* "Elmarikh", *Accia tortilis sub spp. raddiana* "Elseial", *Zizphus spinichristi* "Elsider", *Adensonia digitata* "Eltabaldi", *Capparis decidua* " Eltundob" and *Acacia senegal* "Elhashab". Grasses include *Cenchrus biflorus, C. setigrus, C. citiatis* and *C. periuri* "Elhaskaniet", *Aristida adscensiois* "ELGao" and *chrosophora broocchia* "Elargosae"

٢,٢,٣ Climate

The annual rainfall reaches averages of ٣٠٠ mm in the North to ٦٠٠ mm in the South of Kordofan. Temperatures range between ١٠°C in the cool dry season (from November, February) to ٤٥°C in the hot dry season (from March, June), while in the hot wet rainy season (from July, October), the temperature varies according to humidity from ٢٥°-٤٥°C (Metrological Authority, Ministry of Aviation (٢٠٠٥)).

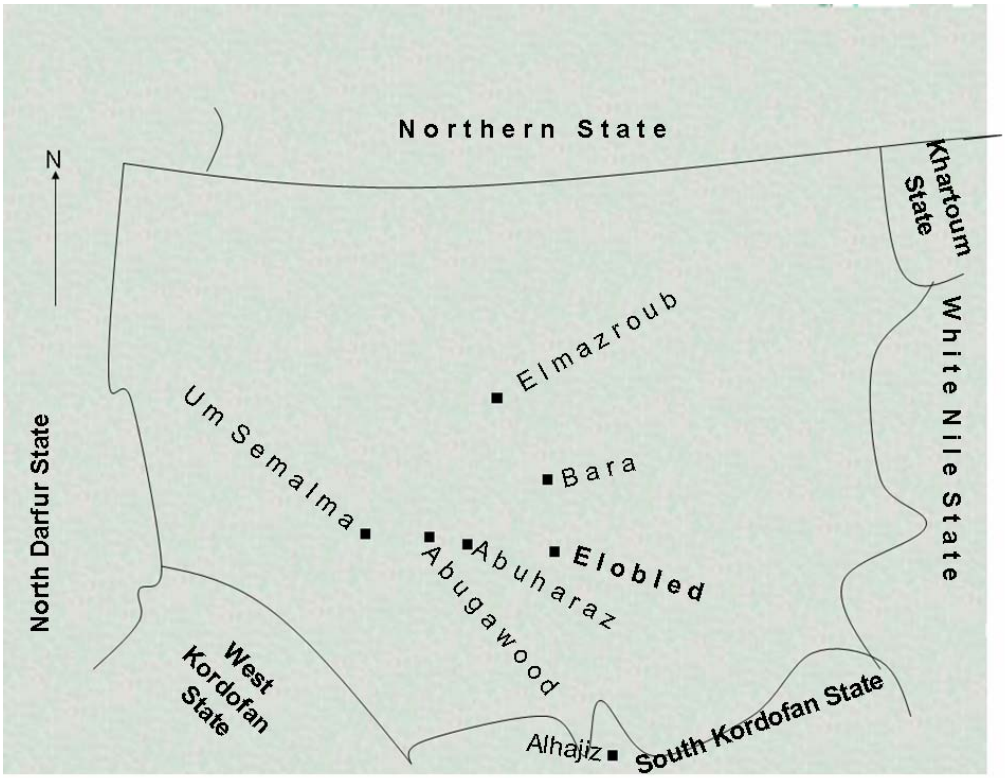


Figure (٢،١): Study area in North Kordofan State

٢،٣. Questionnaire survey

The general questionnaire survey for camel mastitis was carried out in North Kordofan State at Elobied livestock market, ElHajiz camel market ١٢٠ km South of Elobied, Elmazroub camel market ١٦٥ km North of Elobied and (Abu haraz, Abu Gawood and um semaima ٤٠, ٤٧, ٦٥ km respectively) west of Elobied. The questionnaire including presence of mastitis, cause of mastitis, treatment of camel mastitis. the use of anti-suckling devices, practices of cauterization as mean of treating udder problems, tick infestations problem, general diseases infected camel (local and scientific name) and methods of treatment (local and systemic treatment). Type of vaccine used, main constraints of camel rearing, the migration routes of nomadic camel movements and methods of their control**.** Attached design of general questionnaire survey.

General questionnaire

Date ( ) Serial No.( )

Owner's name( ) Area of study ( )

Education level ( ) Tribe ( )

١- Veterinary and extension services: Yes ( ) No ( )

٢- The main constraints of camels rearing:

Feed shortage ( ) Water scarcity ( ) Disease ( )

٣- Presence of mastitis: Yes ( ) No. ( )

٤- Cause of camel mastitis: ( )

٥- Treatment of camel mastitis: Local ( ) Systematic ( )

٦- Application of cauterization for udder problems: Yes ( ) No ( )

٧- Use of anti-suckling devices: Yes ( ) No ( )

٨- Tick control: Manual removal ( ) Use of acaricides ( )

Others( )

٩- Frequency of milking :Once ( ) Twice ( ) Three ( )

Four ( )

١٠- Cleaning the udder before milking: Yes ( ) No ( )

١١- Washing the hands before milking **:** Yes ( ) No ( )

١٢- Migration routes of camel ( )

١٣- Locality of camel stay : Summer ( )

Winter ( ) Autumn ( )

١٤- Other disease problem (

)

١٥- Treatment by whom (

)

١٦- Vaccination if any ( )

Type ( ) Season ( ) Source ( )

١٧- Camel calf problem ( )

٢,٤. Samples for bacteriological analysis

٢,٤,١ Collection of quarter milk samples

Collection of samples were done according to availability of camel herds as well as acceptance of the camel owners (non probability sampling method. Thrusfied, ١٩٩٥). Atotal of ٢١٦ milk samples from ٦٠ lactating she-camels from different parts of North Kordofan State (Elobied, Bara, Abu gawood, Abu haraz and um semaima) were collected. Most herds were owned by nomads except one flock at Kordofan University which is kept on semi closed system for experimental study. Before the collection of quarter milk samples, the suckling calf was used to stimulate milk let-down, the teats were disinfected with cotton wool moistened with ٧٠٪ ethyl alcohol. The first few squirts of milk were discarded and about ٥-١٠ ml of milk were collected in a sterile disposable plastic tube. The quarter milk samples were kept in an ice-box and transported as soon as possible to Elobied Veterinary research laboratory and were cultured. White side test and a slide for Somatic Cell Counts (SCC) were prepared from each sample. After that, the samples were kept at -٢٠°C. The questionnaire for sample collection were based on breed, age, stage of lactation, number of calving, previous history of mastitis and abnormalities of the udder and milk. Attached design of sampling questionnaire collection.

Questionnaire of sample collection

Date of sample collection ( ) Serial No ( )

Owner's name( )Area of study( )

١. Animal identification : Age( ) Calving date( )

٢. Stage of lactation ( )

٣. Number of calving.( )

٤. Previous history of udder problem: Yes ( ) No ( )

٥. Physical examination of udder:

Conformation: Even ( ) Uneven ( )

٦. Teat lesion: Present ( ) Absent ( )

٧. Tick infestation :Present ( ) Absent ( )

٨. Cross milk quality:

Watery ( ) Blood tinget ( ) Clots/flak's ( ) Normal ( )

٩. W.S.T. :

RF* ( ) RB ( ) LF( ) LB( )

١٠.S.C.C. :

RF ( ) RB ( ) LF( ) LB( )

١١.Form of mastitis

١٢. Clinical ( ) Sub-clinical ( )

(RF* = Right front RB= Right behind LF= Left front LB= Left behind) = Quarter of udder

٢,٤,٢ Examination of quarter milk sample

٢,٤,٢,١ White Side Test (WST)

The white side test was performed according to Murphy and Hanson (١٩٤١) on glass slide onto black ground, ٤٪ NaOH was mixed with milk of each quarter in a ratio of ١:٥ according to the degree of milk thickening the test was scored. Normal milk showed no change while mastitic milk become viscid and thick.

٢,٤,٢,٢ Somatic Cell Count (SCC)

The direct microscopic somatic cell count (D.M.S.C.C.) was applied as recommended by Packarrd *et al* (١٩٩٢). The following procedure was employed, an amount of (٠,٠١ ml milk sample was spread over an area of ١ $cm^{٢}$ on a glass slide. The smear was dried and heated slowly to prevent cracking and peeling. The smears were stained with Newman's stain for two minutes, wash gently in water, and then the average count is multiplied by the Microscopic Factor (MF).

$$\text{Leucocyte count} = \frac{\text{Number of leucocytes counted} \times MF}{\text{Number of fields counted}}$$

$$\text{Magnification factor (MF)} = \frac{٤٠٠٠٠}{٣,١٤١٦ \times d^{٢}}$$ where d = diameter

The somatic cell counts (SCC) of ٢,٥×١٠^{٥} cell ml $^{-١}$ was used as the cut-off point (Radostits *et al*., ٢٠٠٠) to classify quarter milk samples of the lactating she-camel as positive in the analysis.

٢,٥ Culture media

٢,٥,١ Preparation of solid media

٢,٥,١,١ Blood agar (Barrow and Feltham, ١٩٩٣)

The medium contained nutrient agar ٩٠٠ ml, sterile defibirinated blood ١٠٠ ml and prepared according to the manufacture's instructions, sterilized by autoclaving at ١٢١°C for ١٥ minutes cooled to ٥٠°C and aseptically sterile blood was added and mixed. The medium was then poured in ٢٠ ml amounts in sterilized petridishes. The pH range of ٧,٢- ٧,٦ at room temperature . The prepared medium was kept at ٤°C until use.

٢,٥,١,٢ Nutrient agar (Oxoid)G/L

The medium contained lab-lemco powder ١ gm, yeast extract ٢ gm, peptone ٥ gm, sodium chlorides ٥ gm and agar No.٣, ١٥ gm. according to the manufactures instructions the medium was prepared by dissolving ٢٨ grams of the powder in one liter of distilled water then sterilized and poured in ٢٠ ml amounts in sterilized petridishes. pH ٧,٤

٢,٥,١,٣ Diagnostic Sensitivity Test (DST) agar (Oxoid) G/L

The medium composed of proteose peptone ١٠ gm, veal infusion solids ١٠ gm, Dextrose gm ٢, Sodium chloride ٣ gm, Disodium phosphate ٢ gm, Sodium acetate ١ gm, Adenine sulphate ٠,٠١ gm, Guanine hydrochloride ٠,٠١ gm, Uracil ٠,٠١ gm, Xanthine ٠,٠١ gm, Aneurine ٠,٠٠٠٢ gm, Ion agar No.٢, ١٢. gm. According

to manufacture's instructions by dissolving ٤٠ grams in one liter of distilled water by boiling, sterilized by autoclaving at ١٢١°C for ١٥ minutes and poured into peteridishes in ١٥ ml portions. Then stored at ٤°C until use.

٢,٦ Preparation of semisolid media

٢,٦,١ Hugh and liefson's (O.F) medium (Barrow and Feltham, ١٩٩٣) G/L

The medium contained Sodium chloride ٥ gm, peptone ٢ gm, agar ٣ gm, K_2HPO_4 ٠,٣ gm, and bromthymol blue (٠,٢٪ aqueous solution) ١٥ml, the ingredients were dissolved by heating in water bath set at ٥٥°C , the pH was adjusted to ٧,١, then the indicator was added and the medium sterilized at ١١٥°C, for ٢٠ minutes, a volume of ١٠ ml of sterile glucose solution was aseptically added to ٩٠ ml of medium. Then the medium was mixed and distributed aseptically in ١٠ ml amounts into sterile test tubes. The prepared medium was kept at ٤°C, until use.

٢,٦,٢ Motility Medium (Barrow and Feltham, ١٩٩٣) G/L

The ingredients are peptone ١٠ gm, meat extract ٣ gm, sodium chloride ٥ gm, agar ٤ gm, Gelatain ٨٠ gm and Distilled water ١٠٠٠. The gelatain was soaked in water for ٣٠ minutes, then the other ingredients were added, heated to dissolve, and sterilized at ١١٥°C for ٢٠ minutes. The prepared medium was kept at ٤°C until use.

٢,٧ Preparation of liquid media

٢,٧,١ Nutrient broth (Oxoid) G/L

The medium contained lab-lemoco powder ١ gm, yeast extract (Oxoid L ٢٠) ٢ gm, peptone (Oxoid L ٣٧) ٥ gm and sodium chloride ٥ gm, according to

manufacture's instructions by dissolving ١٣ grams of the powder in one liter of distilled water and sterilize. Then distributed aseptically in ١٠ ml volumes in sterile tubes. pH ٧,٢- ٧,٤.

٢,٧,٢ Nitrate broth medium (Barrow and Feltham,١٩٩٣) **G/L**

The medium contained Potassium nitrate ١٠ gm, Nutrient broth ١٠٠٠ml. According to the manufacture's instructions the medium was prepared by dissolving one gram potassium nitrate in ١٠٠ ml nutrient broth. Then distributed in ٥ ml amounts in test tubes and sterilized by autoclaving.

٢,٧,٣ Peptone water (Oxiod) G/L

This medium contained peptone ١٠ gm, sodium chloride ٥ gm, it was prepare according to Cowan and Steel (١٩٨٥) by dissolving ١٠ grams of peptone and ٥ grams of sodium chloride in one liter of distilled water. The medium was distributed in ٥ ml amounts in Mc Cortany bottles and sterilized. pH ٧,٢.

٢,٧,٤ Peptone water sugars (Barrow and Feltham, ١٩٩٣)

This medium contained peptone water ٩٠٠ ml. ١٠ ml andrade's indicator. The pH of peptone water was adjusted to ٧,١- ٧,٣ before the andrade's indicator was added, then the specific sugar was added and mixed, then distributed in portions of five ml into sterile test tubes and sterilized by autoclaving at ١٠ pressure for ١٠ minutes, then stored at ٤°C until use.

٢,٧,٥ Carbohydrate fermentation media

These were prepared according to Cowan and Steel (١٩٨٥) ٩٠٠ml of peptone water was prepared it is pH was adjusted to ٧,٢- ٧,٤, ١٠ ml of bromocresol purple

was added and the medium was then sterilized. The colour of the medium is purple. The sugar solution was prepared by dissolving ١٠ grams of sugar in ٩٠ ml of sterilized distilled water. This amount was added aseptically to the sterile peptone water and indicator. The medium was distributed in ٥ ml amounts in sterile test tubes with inverted Durham tube and sterilized.

٢,٧,٦ MR-VP medium (Oxoid) G/L

This medium contained peptone ٥ gm, Dextrose ٥ gm, and phosphate buffer ٥ gm. It was prepared according to Cowan and Steel, (١٩٨٥) by dissolving in one liter of distilled water and the pH was adjusted to ٧,٥, Glucose was then added to give ٠,٥٪ concentration. The medium was distributed in ٥ml amounts in test tubes and sterilized.

٢,٨ Reagents and solutions

٢,٨,١ Sodium hydroxide

This was obtained from (B.D.H.). It was prepared according to Cowan and Steel (١٩٨٥) as ٤٪ solution by dissolving ٤ grams of pure sodium hydroxide in ١٠٠ ml of distilled water. It was used for the white side test.

٢,٨,٢ Nitrate reagent

٢,٨,٢,١ Solution (A)

This was prepared by dissolving ٠,٨٪ sulphanilic acid (Hopkin and Williams) in ٥/N acetic acid.

٢,٨,٢,٢ Solution (B)

This was prepared by dissolving ٠,٥٪ ١- naphthylamine in ٥/N acetic acid. They used for nitrate reduction.

٢,٨,٣ Hydrogen peroxide (H_2O_2)

This was prepared as ٣٪ aqueous solution for the catalase test.

٢,٨,٤. Tetramethyl-P-phenylene diamine dihydrochloride

This was obtained from Hopkin and willian (London it was prepared as ١٪ aqueous solution and used for the oxidase test.

٢,٨,٥ Potassium hydroxide (Barrow and Feltham, ١٩٩٣)

This was obtained from (B.D.H.) and prepared as ٤٠٪ aqueous solution for V.P test.

٢,٨,٦ α-Naphthol solution (Barrow and Feltham, ١٩٩٣)

This reagent was obtained from (B.D.H.) and prepared as ٤٥ aqueous solution for voges-proskaur test.

٢,٨,٧ Methyl red test

This was prepared according to Cowan and Steel (١٩٨٥), by dissolving ٠,٠٤ grams methyl red in ٤٠ml ethanol and volume was made to ١٠٠ ml in distilled water. It was used for V.P test.

٢,٩ Indicators

٢,٩,١ Bromocresol purple

It was prepared according to Cowan and Steel (١٩٨٥), by dissolving ٠,٢ grams of powder in ١٠٠ ml distilled water. It was supplied by (B.D.H.).

٢,٩,٢ Bromothymol blue

This indicator was obtained from (B.D.H.) and prepared by dissolving ٠,٢ gram powder in ١٠٠ ml distilled water.

٢,٩,٣ Andrade's indicator (Barrow and Feltham, ١٩٩٣)

Five grams of acid fuchsin was dissolved in one liter of the distilled water then ١٥٠ ml of N-NaOH was added, mixed and left at room temperature for ٢٤ hours, the colour should change from red to brown.

٢,٩,٤ Human and rabbit plasma

It was used for coagulase test.

٢,١٠ Culture methods

٢,١٠,١ Primary isolation

٢,١٠,١,١ Solid media

All white side test positive and negative samples were cultured by streaking a full wireloop onto solid media and incubated at ٣٧°C for ١٨-٢٤ hours.

٢,١٠,١,٢ Sub-culturing of primary isolate

٢,١٠,١,٢,١ From solid to solid media

One colony was picked with a wireloop and then streaking into the corresponding solid media and incubated at ٣٧°C for ١٨-٢٤ hours.

٢,١٠,١,٢,٢ From solid to liquid media

One colony was picked with a wireloop and then dipped onto the corresponding liquid media and incubated.

٢,١١ Incubation of cultures

Solid and liquid media were incubated aerobically at ٣٧°C for ١٨-٢٤ hours. MR-VP and peptone water for indol test were incubated for ٤٨ hours.

٢,١٢ Examination of cultures

Culture on solid media were examined after ١٨-٢٤ hours of inoculation with the naked eye for growth, colonial morphology and haemolytic characteristics. Liquid media were examined for turbidity, change in colour, formation of sediments and accumulation of gas in the Durham's tube in case of carbohydrate media.

٢,١٣ Purification and preservation of cultures

Pure cultures were obtain by subclturing part of typical and well isolated colony on crossponding medium. This method was repeated at least twice. The resulting growth was checked for purity by examining smears stained by Gram's methods. Pure cultures were stored at -٢٠°C until use for different tests.

٢,١٤ Staining technique

٢,١٤,١ Preparation of smears

٢,١٤,١,١ For bacteria

Smears were prepared by emulsifying and spreading part of a colony or spreading a loopful of culture on a clean slide, the smears were allowed to dry by air and then fixed by gentle flaming and stained.

٢,١٤,١,٢ For leukocyte count

Smears were prepared by taken ٠,٠١ ml of milk and spread over an area of ١ $cm^{٢}$ of a glass slide. Then allow to dry in air.

٢,١٥ Staining methods

٢,١٥,١ Gram's stain

According to Cowan and Steel (١٩٨٥). It was done as follows:

١. Crystal violet was added to smear for one minute
٢. Wash with water and flood with lugol's iodine for one minute
٣. Wash with water
٤. Decolorized with alcohol for ٢-٣ seconds
٥. Wash with water
٦. Counter stain with diluted Carbol fuchsin for one minute
٧. Wash with water
٨. Dry with filter paper and examine under oil immersion
٩. Gram positive organisms appear purple while gram negative ones appear red.

٢,١٥,٢ Newman's stain

This was done according to Cowan and Steel (١٩٨٥)

١. Allow slide of milk to dry in air. .
٢. Heat slowly to prevent cracking and peeling

٣. Immerse smear for about one minute in Newman's stain

٤. Wash gently in water and dry in air

٥. Examine stained smear under oil immersion

٦. Count the leukocyte in at least twenty fields and take the average. Then the average is multiplied by the microscopic factor (MF)

$$\text{Leukocyte count} = \frac{\text{Number of leukocyte counted}}{\text{Number of fields counted}} \text{ MF}$$

٢,١٦ Identification of bacteria

The purified isolates were identified according to Cowan and Steel (١٩٨٥) as follows:

١. Reaction to gram's stain

٢. Shape of organisms

٣. Presence or absence of spore

٤. Motility

٥. Aerobic growth

٦. The colonial characteristics on the different media and haemolysis

٧. Biochemical tests

The biochemical tests were performed according to Cowan and Steel (١٩٨٥). They include:

٢,١٦ .١ Catalase test

On clean microscopic slide, aloopful of ٣٪ hydrogen peroxide was placed. Isolated colony on a plate of nutrient agar was picked with a thin glass rod and put in the reagent. Production of gas bubbles indicated positive result.

٢,١٦,٢ Oxidase test

Pieces of filter paper were soaked in ١٪ solution of tetramethylene-P-phenylene diamine dihydrochloride and dried. A colony on nutrient agar was picked with sterile bend glass rod and smeared across the surface of filter paper in a Petridish. A dark purple colour within ١٠ seconds was considered as positive reaction (Cruick shank, ١٩٧٥).

٢,١٦,٣ Oxidation fermentation (O/F) test

Tow tubes of Hugh and leifson's medium were inoculated with the test organism by stabbing with straight wire. One of the tubes was covered by alayer of sterile paraffin oil of one Cm. above the surface of the medium, the other was left uncovered. Tubes were incubated and examined daily for tow weeks. If colour change to yellow in both open and sealed tubes this indicated fermentative organism, but change in the uncovered tubes indicated that the organism was oxidative.

٢,١٦,٤ Motility test

A small piece of the colony of bacterium under test was picked by the end of the straight wire and stabbed in the center of the semisolid agar in the inner side of craigie tube. This preparation was incubated at ٣٧°C overnight the organism was

considered motile if there was turbidity in the medium in and outside the craigie tube.

٢,١٦,٥ Sugars Fermentation Test

The test organism was inoculated into peptone water sugar medium, incubated at ٣٧°C and examined daily for up to ٧ days. Acid production was indicated by the development of colour in medium. While gas production was shown by development of an empty space in Durham tube.

٢,١٦,٦ Urease Test

Aslope of urea agar medium was inoculated with the test organism, incubated and examined after ٢٤ hours and daily for ٥ days. Change of colour to red indicated positive reaction.

٢,١٦,٧ Nitrate reduction

Nitrate broth was inoculated with the test organism and incubated for ٤٨ hours, after incubation, one ml of nitrate solution (A) followed by one ml of nitrate solution (B) were added. Red colour indicated positive reaction, tube that don't show red colour within ٥ minutes, powdered Zinc up to ٥ mg/ml of cultures was added. Red colour indicated positive reaction.

٢,١٦,٨ Voges-proskauer (V.P) reaction

Glucose phosphate medium was inoculated with the test organisms and incubated for ٤٨ hours. ٠,٦ ml of ٥٪ α-naphthol followed by ٠,٢ ml of ٤٠٪ potassium

hydroxide aqueous solution were added. It was shaken and examined after ١٥ minutes and one hour. A positive reaction was indicated by strong red colour.

٢,١٦,٩ Coagulase test

One ml of plasma was added to nine ml normal saline to make ١:١٠ dilution. Then ٠,٥ ml of this dilution was mixed with ٠,٥ ml of broth culture in agglutination tube, incubated at ٣٧°C and examined after four hours. The positive result was indicated by definite clot. Negative tubes showed no clot formation when examined after overnight incubation at ٣٧°C.

٢,١٦,١٠ CAMP test (Quinn, ١٩٩٤)

A culture of β-haemolysis *staphylococcus aureus* was streaked across the centre of sheep blood agar plate. The suspect organism was made at right angles to, and taken to within ١,٥ mm of *staphylococcus* streak. The plate was incubated over night at ٣٧°C.Apositive CAMP test was indicated by an arrow-head of complete haemolysis.

٢,١٦,١١ Novobiocin sensitivity test

A volume of two ml of diluted culture were spreads on the surface of nutrient agar. The excess fluid was discarded and the plate was allowed to dry, then Oxoid discs of novobiocin (٥mg) was applied to surface of medium by sterile forceps and

incubated at ٣٧°C for ٢٤ hours. Zone of inhibition was determined whether the organism was sensitive or not to novobiocin.

٢,١٦,١٢ Antibiotic sensitivity test

The test was performed by the standard disc diffusion method (Cruickshank *et al*, ١٩٧٥). The organisms were sub-cultured onto blood agar and incubated at ٣٧°C for ١٨-٢٤hrs. they were diluted in ٤ml sterile normal saline tubes and ٢ml of diluted cultures were spread onto surface of nutrient agar and D.S.T. agar after drying. Excess fluid was aspirated by sterile pipette, and the plates were allowed to dry. Oxoid discs (Basingtok, Hampshire, England) of antimicrobial drugs were applied to the surface of the medium and pressed gently using sterile forceps. They were incubated at ٣٧°C for ٢٤-٤٨ hrs. zones of inhibition were measured in (mm) to determine whether the organism was sensitive or resistant (Jacoby and Archer, ١٩٩١).

Table (٢،١): Zone diameter interpretive chart: (WHO, ١٩٧٧).

Antimicrobial agent	Disk contents	Zone of inhibition (Diameter) in mm		
		Resistant ≤	Intermediate	Sensitive ≥
Ampicillin Am	١٠ mcg	٢٢	٢٣-٣٠	٣١
Cephalexine CFX	٣٠ mcg	١٤	١٥-١٧	١٨
Ciprofloxacin CIP	٥ mcg	١٥	١٦-٢٠	٢١
Cefotaxime CTX	٣٠ mcg	١٤	١٥-٢٢	٢٣
Co.Trimoxazole SXT	٢٥ mcg	١١	١٢-١٦	١٧
Gentamycin GN	١٠ mcg	١٢	١٣-١٤	١٥
Lincomycin LN	٢ mcg	٩	١٠-١٤	١٥
Offloxacin OFF	٥ mcg	١٥	١٦-٢٠	٢١
Cloxacinllin CLX	٥ mcg	٩	١٠-١٣	١٤
Tetracycilin TE	٣٠mcg	١٤	١٥-١٨	١٩

CHAPTER THREE

RESULTS

٣,١ Questionnaire survey

The investigation was carried out to give information about mastitis, predisposing factors and other problems associated with the disease in North Kordofan State. About ١١٨ camel's owners were interviewed (٢٩, ١٥ and ٧٤) in the Elobied, ElHajiz and Emazroub respectively. The data from general questionnaire survey are summarized in table (٣,١).

The results showed that, (٨٥,٥٩٪) of camel owners do not have veterinary and extension services. (٥٥,٠٨٪) confirmed the major constrains of camel rearing are feed shortage and water scarcity. Where as, (٣٧٪) said that, the problem was diseases. (٨٣,٠٥٪) confirmed presence of anti-suckling devices without milking is the main causes of mastitis but only ٤,٢٤٪ and ٧,٦٣٪ due to ticks and flies respectively, as the major causes of mastitis. (٩٦,٦١٪) used the anti-suckling devices is known by owners “surar” to keep milk for themselves, which is the strong predisposing factor for mastitis. However, (٨٥,٥٪) said that, acaricides used for tick control were responsible for this problem. While ١٣,٥٦٪ use “Gutran” for tick control. (٩٢٪) of owners do not wash their hands before milking.

On the other hand, the results of sampling questionnaire showed that. ١٠٠٪ of lactating she-camel were infested with tick, ٧٨,٣٣٪ had even udder and ٥٦,٦٧٪ had teat lesion. The results of sampling questionnaire are shown in table (٣,٢).

Table(٣،١) Result of questionnaire survey collected from camels owners in North Kordofan State

Factors	El Obeid N(%)	El Hajiz N(%)	El Mazroub N(%)	Total N(%)
Constrains of camel rearin				
١. Feed shortage and water scarcity	١٧(٥٨,٦٢)	١(٦,٦٧)	٤٧(٦٣,٥١)	٦٥(٥٥,٠٨)
٢. Diseases	١١(٣٧,٩٢)	١٢(٨٠,٠٠)	١٤(١٨,٩٢)	٣٧(٣١,٣٦)
٣. Both	٠(٠,٠٠)	١(٦,٦٧)	١٣(١٧,٥٧)	١٤(١١,٨٦)
٤. Pasture and insecurity	١(٣,٤٥)	١(٦,٦٧)	٠(٠,٠٠)	٢(١,٦٩)
Veterinary and extension services				
١. Yes	٤(١٣,٧٩)	٥(٣٣,٣٣)	٨(١٠,٨١)	١٧(١٤,٤١)
٢. No	٢٥(٨٦,٢١)	١٠(٦٦,٦٧)	٦٦(٨٩,١٩)	١٠١(٨٥,٥٩)
Presence of mastitis				
١. Yes	٢٩(١٠٠)	١٥(١٠٠)	٧٣(٩٨,٦٥)	١١٧(٩٩,١٥)
٢. No	٠(٠,٠٠)	٠(٠,٠٠)	١(١,٣٥)	١(٠,٨٥)
Cause of mastitis				
١. Tick	٠(٠,٠٠)	٤(٢٦,٦٧)	١(١,٣٥)	٥(٤,٢٤)
٢. Flies	١(٣,٤٥)	١(٦,٦٧)	٧(٩,٤٦)	٩(٧,٦٣)
٣. Both	٠(٠,٠٠)	٢(١٣,٣٣)	٢(٢,٧٠)	٤(٣,٣٩)
٤. Presence of anti-suckling device without milking	٢٨(٩٦,٥٥)	٧(٤٦,٦٧)	٦٣(٨٥,١٤)	٩٨(٨٣,٠٥)
٥. Nothing	٠(٠,٠٠)	١(٦,٦٧)	٠(٠,٠٠)	١(٠,٨٥)
Type of treatment				
١. Antibiotic	١٩(٦٥,٥٢)	٧(٤٦,٦٧)	٤٢(٥٦,٧٦)	٦٨(٥٧,٦٢)
٢. Traditional	٣(١٠,٣٤)	٤(٢٦,٦٧)	١٧(٢٢,٩٧)	٢٤(٢٠,٣٤)
٣. Both	٦(٢٠,٦٩)	٣(٢٠,٠٠)	٦(٨,١١)	١٥(١٢,٧١)
٤. Nothing	١(٣,٤٥)	١(٦,٦٧)	٨(١٠,٨١)	١٠(٨,٤٧)
Practice of cauterization				
١. Yes	٢(٦,٩٠)	٥(٣٣,٣٣)	٩(١٢,١٦)	١٦(١٣,٥٦)
٢. No	٢٧(٩٣,١٠)	١٠(٦٦,٦٧)	٦٤(٨٦,٤٢)	١٠١(٨٥,٥٩)
Use of anti-suckling devices				
١. Yes	٢٨(٩٦,٥٥)	١٤(٩٣,٣٣)	٧٢(٩٧,٣٠)	١١٤(٩٦,٦١)
٢. No	١(٣,٤٥)	١(٦,٦٧)	٢(٢,٧٠)	٤(٣,٣٩)
Tick control				
١. Acaricides	٢٩(١٠٠)	١٤(٩٣,٣٣)	٥٨(٧٨,٣٨)	١٠١(٨٥,٥٩)
٢. Manual removal	٠(٠,٠٠)	١(٦,٦٧)	٠(٠,٠٠)	١(٠,٨٥)
٣. Both	٠(٠,٠٠)	٠(٠,٠٠)	٠(٠,٠٠)	٠(٠,٠٠)
٤. *"Gutran"**	٠(٠,٠٠)	٠(٠,٠٠)	١٦(٢١,٦٢)	١٦(١٣,٥٦)

Frequency of milking				
١. Once	٢(٦,٩٠)	١(٦,٦٧)	١(١,٣٥)	٤(٣,٣٩)
٢. Twice	١٠(٣٤,٤٨)	٧(٤٦,٦٧)	٤٣(٥٨,١١)	٦٠(٥٠,٨٥)
٣. Three times	١٤(٤٨,٢٨)	٥(٣٣,٣٣)	٢٨(٣٧,٨٤)	٤٧(٣٩,٨٣)
٤. Four times	٣(١٠,٣٤)	٢(١٣,٣٣)	٢(٢,٧٠)	٧(٥,٩٣)
Cleaning of the udder before milking				
١. Yes	١(٣,٤٥)	١(٦,٦٧)	١(١,٣٥)	٣(٢,٥٤)
٢. No	٢٨(٩٦,٥٥)	١٤(٩٣,٣٣)	٧٣(٩٨,٦٥)	١١٥(٩٧,٤٦)
Washing the hands before milking				
١. Yes	١(٣,٤٥)	٤(٢٦,٦٧)	٤(٥,٤١)	٩(٧,٦٣)
٢. No	٢٨(٩٦,٥٥)	١١(٧٣,٣٣)	٧٠(٩٤,٥٩)	١٠٩(٩٢,٣٧)

*Gutran** = traditional material used for tick control

N= number (%) percentage

Table (٣،٢) Result of sampling questionnaire, some information collected from the camel owners and the others by visual examination

Factors	West of Elobeid N(%)	Elobeid N(%)	Bara N(%)	Total N(%)
Tick infestestion				
Present	٤١(١٠٠)	٨(١٠٠,٠)	١١(١٠٠,٠)	٦٠(١٠٠,٠)
Absence	٠(٠٠,٠)	٠(٠٠,٠)	٠(٠٠,٠)	٠(٠٠,٠)
Conformation of udder				
Even	٣١(٧٥,٦١)	٧(٨٧,٥)	٩(٨١,٨٢)	٤٧(٧٨,٣٣)
Uneven	١٠(٢٤,٣٩)	١(١٢,٥)	٢(١٨,١٨)	١٣(٢١,٦٧)
Teat lesion				
Presence	٢١(٥١,٢٢)	٣(٣٧,٥)	١٠(٩٠,٩١)	٣٤(٥٦,٦٧)
Absence	٢٠(٤٨,٧٨)	٥(٦٢,٥)	١(٩,٠٩)	٢٦(٤٣,٣٣)
Stage of lactation				
First	٥(١٢,٢٠)	١(١٢,٥)	٣(٢٧,٢٧)	٩(١٥,٠٠)
Second	١١(٢٦,٨٣)	٥(٦٢,٥)	٠(٠٠,٠٠)	١٦(٢٦,٦٧)
Third	١٧(٤١,٤٦)	٢(٢٥,٠)	٧(٦٣,٦٤)	٢٦(٤٣,٣٣)
Fourth	٨(١٩,٥١)	٠(٠٠,٠)	١(٩,٠٩)	٩(١٥,٠٠)
Age				
٠-٥ year	٦(١٤-٦٣)	٠(٠٠,٠)	١(٩,٠٩)	٧(١١,٦٧)
٦-١٠ year	١٣(٣١-٧١)	٢(٢٥,٠)	٨(٧٢,٧٣)	٢٣(٣٨,٣٣)
> ١٠ year	٢٢(٥٣-٦٦)	٦(٧٥,٠)	٢(١٨,١٨)	٣٠(٥٠,٠٠)
No.of calving				
one year	٨(١٩,٥١)	٠(٠٠,٠)	٣(٢٧,٢٧)	١١(١٨,٣٣)
Two year	٩(٢١,٩٥)	٢(٢٥,٠)	١(٩,٠٩)	١٢(٢٠,٠٠)
Three year	١١(٢٦,٨٣)	٢(٢٥,٠)	٤(٣٦,٢٦)	١٧(٢٨,٣٣)
Four year	١٣(٣١,٧١)	٤(٥٠,٠)	٣(٢٧,٢٧)	٢٠(٣٣,٣٣)
Previous history of udder problems				
Yes	٢٤(٥٨,٥٤)	٣(٣٧,٥)	٣(٢٧,٢٧)	٣٠(٥٠,٠)
No	١٧(٤١,٤٦)	٥(٦٢,٥)	٨(٧٢,٧٣)	٣٠(٥٠,٠)

٣,٢ Results of laboratory tests

٣,٢,١ White Side Test (WST)

A total of ٢١٦ quarter milk samples were examined by (W.S.T.), ٣٥(١٦,٥٠٪) were positive, where as ١٨١ (٨٣,٨٪) were negative. Results are shown in table (٣,٣).

٣,٢,٢ Somatic Cell Count (SCC)

A total of ٢٠٩ quarter milk samples were examined for (S.C.C.), ٣٩(١٨,٦٦٪) were of readings above ٢٥٠،٠٠٠ cells/ml. But ١٧٠ (٨١,٣٤) were below that level. Results are shown in table (٣,٣).

٣,٢,٣ Bacteria isolated from camels mastitis cases

Out of the total number of ٢١٦ quarters milk samples collected from ٦٠ lactating she-camel and cultured for bacteria, ٩٩ (٤٥,٨٣٪) are positive for bacterial growth, where as ١١٧ (٥٤,١٧٪) were negative for bacterial isolation. Results are shown in table (٣,٣). Out of the ٩٩ (٤٥,٨٣) positive cultures, ٦٦ (٣٠,٥٦٪) were purified and identified and the main isolates were *Staphylococci spp*. ٨٠,٣٠٪, *Bacillus spp.* ٩,٠٩٪ and *Corynebacterium spp*. ٣,٠٣٪. Results are shown in table (٣,٤). The cultural and biochemical characteristics used to classify the different types of isolates are shown in tables (٣,٥), (٣,٦), (٣,٧), (٣,٨) and (٣,٩). (Quinn *et al*., ١٩٩٤)

٣,٣ Prevalence of clinical and sub-clinical mastitis

Only quarter milk samples were collected from West of Elobeid gave positive results (٠,٩٣٪) which were positive samples for clinical mastitis cases. Where as

sub-clinical mastitis recorded in all areas of study (West of Elobeid, Elobeid and Bara, ٣٠,٥٩٪, ٥,٥٦٪ and ٩,٧٢٪ respectively). Results are shown in table(٣,١٠).

Table (٣,٣): Summary of the results of white side test, somatic cell count, bacteriological examination and area of collection samples

Unit	Quarters milk sample				
	LB	LF	RB	RF	Total N(%)
	(N/%)				
Area					
West of Elobeid*	٣٨ (٦٩,٠٩)	٣٨(٧٠,٣٧)	٣٧(٦٩,٨١)	٣٨(٧٠,٣٧)	١٥١(٦٩,٩١)
Elobeid	٧ (١٢,٧٣)	٦(١١,١١)	٦(١١,٣٢)	٥ (٩,٢٦)	٢٤(١١,١١)
Bara	١٠ (١٨,١٨)	١٠(١٨,٥٢)	١٠(١٨,٨٧)	١١(٢٠,٣٧)	٤١(٤٨,٩٨)
Sub-total	٥٥(١٠٠,٠٠)	٤٥(١٠٠,٠٠)	٥٣(١٠٠,٠٠)	٥٤(١٠٠,٠٠)	٢١٦(١٠٠,٠٠)
Whit Side Test (WST)					
Positive	٨(١٤,٥٥)	٩(١٦,٦٧)	٩(١٦,٩٨)	٩(١٦,٦٧)	٣٥(١٦,٢٠)
Negative	٤٧(٨٥,٤٥)	٤٥(٨٣,٣٣)	٤٤(٨٣,٠٢)	٤٥(٨٣,٣٣)	١٨١(٨٣,٨٠)
Sub-total	٥٥(١٠٠,٠٠)	٥٤(١٠٠,٠٠)	٥٣(١٠٠,٠٠)	٥٤(١٠٠,٠٠)	٢١٦(١٠٠,٠٠)
Somatic cell count (SCC)					
Positive	٣(٢٤,٠٧)	٦(١١,٣٢)	١١(٢١,٥٧)	٩(١٧,٦٥)	٣٩(١٨,٦٦)
Negative	٤١(٧٥,٩٣)	٤٧(٨٨,٦٨)	٤٠(٧٨,٤٣)	٤٢(٨٢,٣٥)	١٧٠(٨١,٣٤)
Sub -total	٥٤(١٠٠,٠٠)	٥٣(١٠٠,٠٠)	٥١(١٠٠,٠٠)	٥١(١٠٠,٠٠)	٢٠٩(١٠٠,٠٠)
Bacteriological examinations					
Positive	٢٧(٤٩,٠٩)	٢٢(٤٠,٧٤)	٢٨(٥٢,٨٣)	٢٢(٤٠,٧٤)	٩٩(٤٥,٨٣)
Negative	٢٨(٥٠,٩١)	٣٢(٥٩,٢٦)	٢٥(٤٧,١٧)	٣٢(٥٩,٢٦)	١١٧(٥٤,١٧)
Sub -total	٥٥(١٠٠,٠٠)	٥٤(١٠٠,٠٠)	٥٣(١٠٠,٠٠)	٥٤(١٠٠,٠٠)	٢١٦(١٠٠,٠٠)

LB= left behind LF= left front RB= right behind RF= right front
N= number (%) percentage west of Ebeid* = (Abu haraz, Abu Gawood and um semuma)

Table (٣,٤) Frequency and percentage of the bacteria isolated from camel mastitis in North Kordofan State

Species	No. of isolates	Clinical mastitis n(%)	Sub-clinical mastitis n(%)	Total n(%)
Staphylococcus aureus	١٥	٠(٠,٠٠)	١٥(٢٣,٤٤)	١٥(٢٢,٧٥)
S. hyicus	٢	٠(٠,٠٠)	٢(٣,١٣)	٢(٣,٠٣)
S. intermedius	٥	٠(٠,٠٠)	٥(٧,٨١)	٥(٧,٥٦)
S. epidermidis	٨	٠(٠,٠٠)	٨(١٢,٥)	٨(١٢,١٢)
S. delphini	٢	٠(٠,٠٠)	٢(٣,١٣)	٢(٣,٠٣)
S. simulans	٤	٠(٠,٠٠)	٤(٥,٦٣)	٤(٦,٠٦)
S. kloosii	٣	٠(٠,٠٠)	٣(٤,٦٩)	٣(٤,٥٥)
S. carnosus	١	٠(٠,٠٠)	١(١,٥٦)	١(١,٥٢)
S. chromogenes	١	٠(٠,٠٠)	١(١,٥٦)	١(١,٥٢)
S. lentus	٣	٠(٠,٠٠)	٣(٤,٦٩)	٣(٤,٥٥)
S. lugdunensis	٢	٠(٠,٠٠)	٢(٣,١٣)	٢(٣,٠٣)
S. sacchrolyticus	١	٠(٠,٠٠)	١(١,٥٦)	١(١,٥٢)
S. saprophyticus	٢	٠(٠,٠٠)	٢(٣,١٣)	٢(٣,٠٣)
S. haemolyticus	٤	٠(٠,٠٠)	٤(٦,٢٥)	٤(٦,٠٦)
Streptococcus dysgalactiae	١	٠(٠,٠٠)	١(١,٥٦)	١(١,٥٢)
Corynebacterium bovis	٢	٠(٠,٠٠)	٢(٣,١٣)	٢(٣,٠٣)
Bacillus cereus	٦	٢(١٠٠)	٤(٦,٢٥)	٦(٩,٠٩)
Pasteurella haemolytica	٤	٠(٠,٠٠)	٤(٦,٢٥)	٤(٦,٠٦)
Total:	٦٦	٢	٦٤	٦٦

%= percentage.

n= Number of isolates.

Table (٣،٥): The result of biochemical reaction for the identification of gram positive bacteria genus level

Genus of bacteria	**Gram stain**	**Motility test**	**Growth in air**	**Catalase test**	**Oxidase test**	**Glucose test**	**O/F test**
Staphylococcus	+ cocci	-	+	+	-	+	F
Streptococcus	+ cocci	-	+	-	-	+	F
Corynebaterium	+ rods	-	+	+	-	+	F
Bacillus	+ rods	-	+	+	-	+	F

- = negative + = positive F = fermentation

Table (٣،٦): The result of biochemical reaction for the identification of gram negative bacteria genus level

Genusof bacteria	**Gram stain**	**Motility** test	**Growth in air**	**Catalase** test	**Oxidase** test	**Glucose** test	**O/F** test
Pasteurella	**- rods**	**-**	+	+	+	+	**F**

- = negative + = positive F = fermentation

Table (٣،٧): Classification of *Corynebacterium* & *Bacillus* species

Species	Garm stain	Catalase test	Oxidase test	Motility test	Haemolysis	CAMP test	Growthon ٩٪Hcl
Corynebacterium bovis	+ rods	+	-	-	-	-	+
Bacillus cereus	+ rods	+	-	-	+	*	*

+ positive - negative * not known

Table (٣،٨): Classification of *Pasteurella* species

Species	**Gram stain**	**Catalase** test	**Oxidase** test	**Motility** test	**Haemolysis**	**Growth on mac. Agar**	**lactose** test
Pasteurella haemolytica	- Rods	+	+	-	+	+	-

+ = positive - = negative

Table (٣،٩): Classification of *Streptococcus* species

Species	Gram stain	Catalase test	Oxidase test	Motility test	Growth on Mac. Agar	Coagulase	Sorbitol test	CAMP test	Haemolysis	Salicin test
Streptococcus dysgalactiae	**+ cocci**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	+	**-**

Table(٣،١٠): Prevalence of clinical and sub-clinical mastitis in North Kordofan state

Source of quarter milk sample	Total of quarter milk sample	Clinical No. of Positive (prevalence)%	Sub-clinical No. of Positive (prevalence)%
West of Elobeid	١٥١	٢ (١,٣٢)	٦٥ (٣٠,٥٩)
Elobeid	٢٤	-	١٢ (٥,٥٦)
Bara	٤١	-	٢١ (٩,٧٢)

Total	٢١٦	٢ (٠,٩٣)	٩٨ (٤٥,٣٧)

٣,٤ The relationship between laboratory tests

٣,٤,١ The relationship between (WST) and somatic cell count (SCC)

There was no relationship between (WST) and (SCC). Chi square (X^2) = ٢,٧٢١, P.value = ٠,٠٩٩. The results are shown in figure (٣,١).

٣,٤,٢ The relationship between bacteriological examinations and Somatic Cell Counts (SCC)

There was strong correlation between bacteriological examinations and (SCC). (X^2) = chi –square =١٥,٠٥٧, P.value= ٠,٠٠٠. Results are shown figure. (٣,٢).

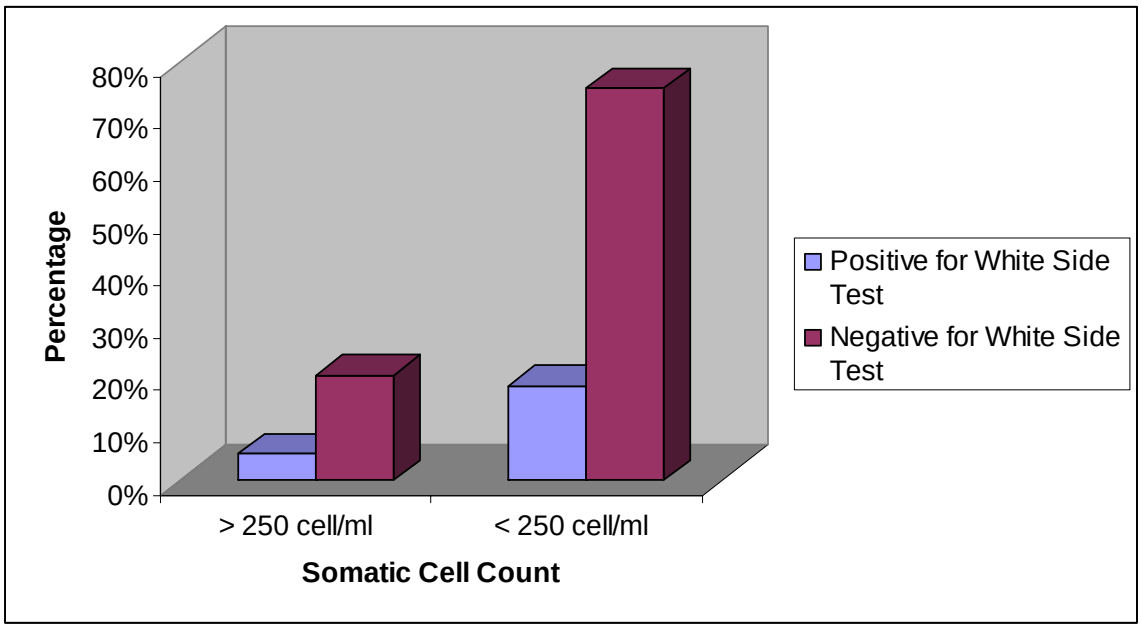
٣,٤,٣ The relationship between White Side Test (WST) and bacteriological examinations

There was strong association between (WST) and bacteriological examinations. Chi-square (X^2) = ٤٤,٢٩٢, P.value= ٠,٠٠٠. Results are shown in figure. (٣,٣).

٣,٥ Result of antibiotic sensitivity test

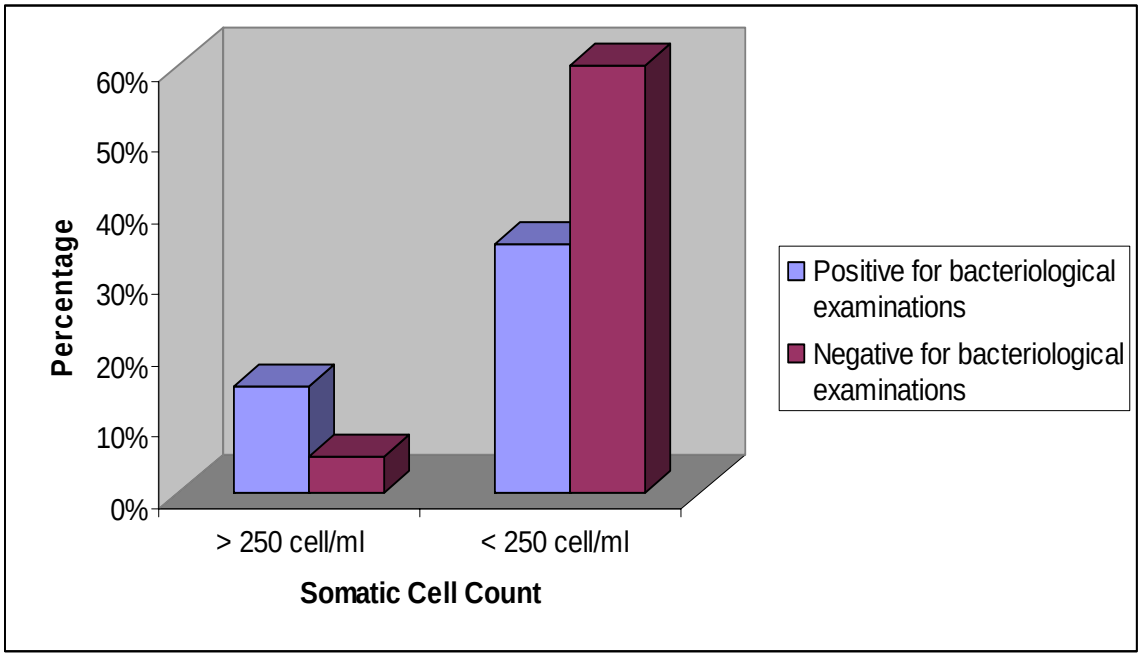
The antibiotic sensitivity test was applied to various isolated bacterial species. The tested organisms could be sensitive or intermediate or resistant to different used antibiotic discs. Our results showed that most of organisms tested were highly sensitive to *Ciprofloxacin, Oflaxacin, Gentamycin, Cephalexine, Tetracycline* and *Co-trimoxazole* but resistant to *Cloxacillin* and *Lincomycin*. However, Ampicillin

and Cefotaxime were considered as causing intermediate inhibition to organisms. These results showed in table (٣،١١).



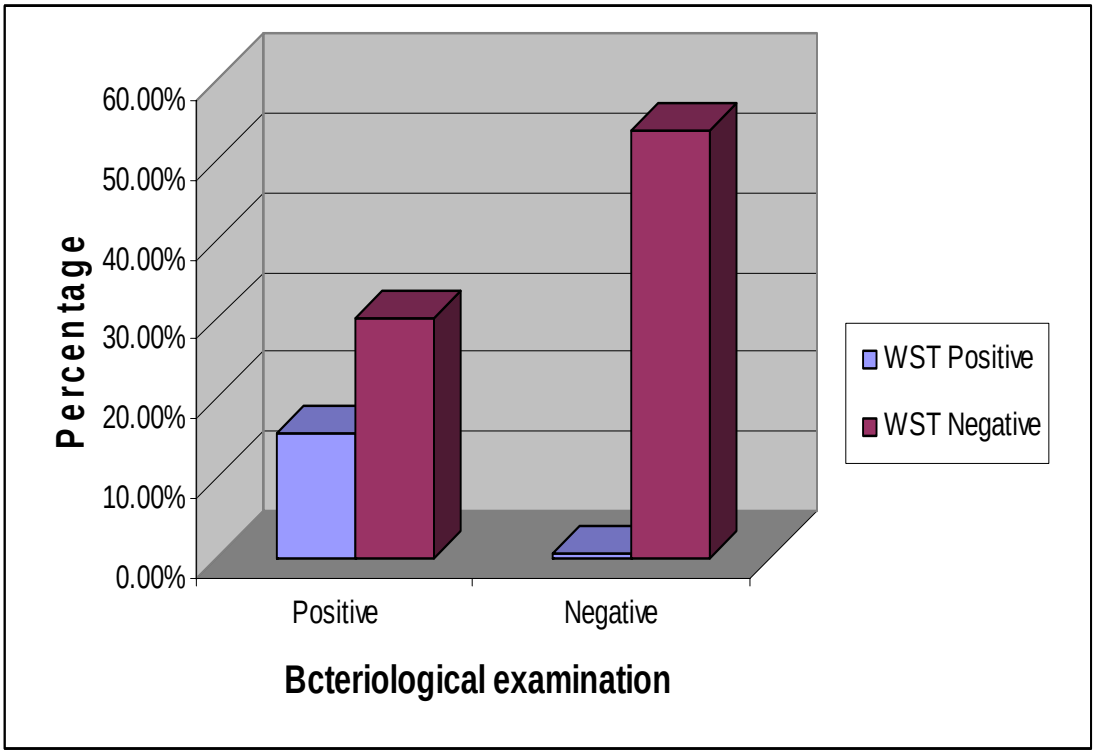
Chi-square (χ^2) = ٢،٧٢١ *P*-value = ٠،٠٩٩ (not significant, $P >$ ٠،٠٥).

Figure (٣،١): The relationship between White Side Test (WST) and Somatic Cell Count (SCC).



Chi-square (χ^2) = ١٥,٠٥٧ *P*-value = ٠,٠٠٠ (highly significant, $P < ٠,٠١$).

Figure(٣,٢): The relationship between bacteriological examinations and Somatic Cell Count (SCC).



Chi-square (χ^2) = ٤٤,٢٩٢ *P*-value = ٠,٠٠٠ (highly significant, $P < ٠,٠١$).

Figure(٣,٣): The relationship between White Side Test (WST) and bacteriological examinations.

Plate (٣،١) The udder heavily infested with ticks

Plate (٣،٢) Injuries on surface of udder and teats associated with mastitis cases

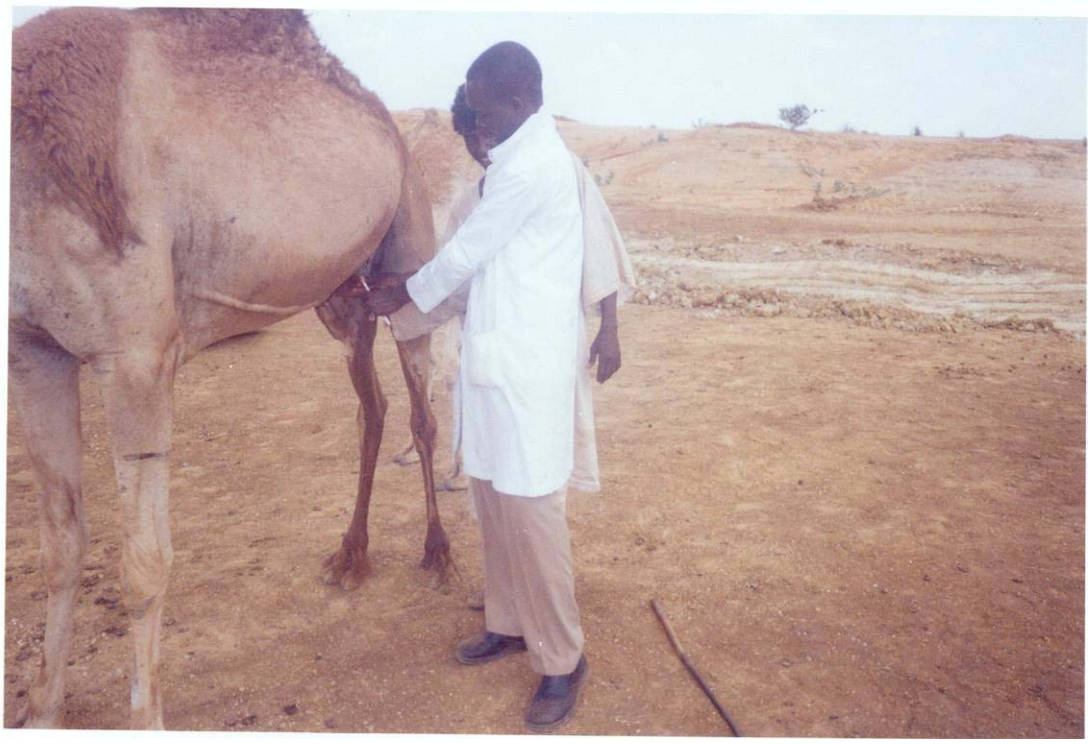


Plate (٣،٣) Method for collection of milk sample in sterile disposable plastic tube

Table (٣،١١): Sensitivity, resistance and intermediate results of various bacterial isolates from she-camel mastitis

Bacterial isolates	AM ١٠ mcg	SXT ٢٥ mcg	CFX ٣٠ mcg	TE ٣٠ mcg	CTX ٣٠ mcg	CIP ٥ mcg	OFF ٥ mcg	CLX ٥ mcg	LN ٢ mcg	GN ١٠ mcg
Staphylococcus aureus	S	R	S	S	Int	Int	Int	R	S	S
S. aureus	Int	R	h^s	S	R	S	S	Int	R	S
S. aureus	Int	h^s	h^s	S	R	S	S	R	R	S
S. aureus	S	R	Int	S	S	S	h^s	Int	Int	S
S. aureus	R	S	Int	S	R	S	S	R	R	h^s
S. aureus	S	h^s	h^s	h^s	R	h^s	S	R	R	S
S. aureus	Int	Int	S	Int	Int	S	Int	R	R	Int
S. aureus	S	R	S	Int	S	Int	S	R	Int	S
S. aureus	S	Int	Int	S	h^s	S	S	Int	R	S
S. hyicus	S	R	S	Int	S	S	Int	Int	R	S
S. hyicus	Int	R	S	S	R	S	Int	R	Int	S
S. intermidius	Int	h^s	S	S	R	S	S	R	R	S
S. intermidius	S	h^s	S	h^s	Int	S	S	Int	S	S
S. epidermidis	S	S	Int	R	Int	R	S	R	S	Int
S. epidermidis	Int	S	S	S	R	h^s	S	R	R	S
S. epidermidis	Int	S	Int	Int	Int	S	S	R	R	S

S. epidermidis	S	Int	Int	Int	Int	S	S	R	Int	S
S. simulans	Int	Int	Int	Intt	Int	S	Int	R	R	Int
S. simulans	S	R	S	S	Int	S	S	Int	Int	S
S. simulans	Int	R	Int	S	Int	S	S	R	R	Int
S. simulans	Int	h^s	h^s	Int	S	h^s	S	Int	Int	h^s
S. kloosii	R	Int	R	Int	R	h^s	S	R	R	S
S. kloosii	R	Int	R	S	R	h^s	S	R	R	S
S. kloosii	R	R	Int	S	R	S	S	R	R	S
S. lentus	S	S	h^s	h^s	h^s	S	S	Int	Int	S
S. lentus	S	S	S	h^s	S	S	S	Int	S	S
S. lentus	Int	R	h^s	S	R	S	S	Int	R	h^s
S. haemolyticus	S	S	S	Int	S	S	S	R	S	S
S. haemolyticus	R	Int	Int	h^s	Int	S	S	R	R	S
S. haemolyticus	Int	h^s	R	S	R	h^s	h^s	R	R	S
S. lugdunensis	S	h^s	h^s	S	Int	S	S	R	S	Int
S. lugdunensis	R	R	S	Int	Int	h^s	S	R	Int	S
S. saprophyticus	h^s	R	h^s	S	Int	S	S	R	Int	S
S. saprophyticus	Int	h^s	h^s	Int	Int	h^s	S	R	R	S
S. delphini	Int	R	S	S	R	h^s	h^s	R	Int	h^s
S. carnosus	R	S	S	Int	Int	S	S	Int	R	S
S. chromogenes	Int	S	h^s	S	Int	h^s	S	R	R	S

S.sacchrolyticus	S	Int	h^s	S	S	S	S	S	R	Int
. *Streptococcus* . *dysgalactiae*	Int	h^s	S	S	R	S	S	R	R	S
Corynebaterium bovis	Int	R	h^s	Int	Int	h^s	S	Int	R	S
Corynebaterium bovis	Int	R	S	S	S	S	S	R	R	S
Bacillus cereus	Int	h^s	h^s	h^s	R	h^s	S	R	R	S
Bacillus cereus	S	S	h^s	S	R	S	S	Int	Int	S
Bacillus cereus	R	Int	Int	S	R	S	Int	R	R	S
Bacillus cereus	S	S	R	Int	Int	S	S	R	R	S
Bacillus cereus	Int	S	Int	S	S	h^s	h^s	S	R	S
Pasteurella haemolytica	S	R	S	S	Int	h^s	S	Int	R	S
Pasteurella haemolytica	S	R	Int	S	S	S	S	R	Int	S

AM= Ampicillin CTX = Cefotaxime LN = Lincomycin TE = Tetracycline

SXT = Co-tromoxazde CIP = Ciproffloxacin GN = Gentamycin CFX = Cephalexine

CLX = Cloxacillin OFF = Ofloxacin

h^s=highlysensitive S = Sensitive INT = Intermediate R = Resistant

CHAPTER FOUR

DISCUSSION

In this Study, the general questionnaire survey showed that out of ١١٨ camel owners interviewed in North Kordofan State, ٩٩٪ of owners confirmed the present of mastitis and ٩٧٪ of them did not clean the udder before milking. This finding agreed with that obtained by Abdel Gadir (٢٠٠١); who reported that ٩٥٪ of tested herds showed the presence of mastitis and ١٠٠٪ did not clean the udder before milking.

On the other hand, ٩٦,٦١٪ of tested herd used the anti-suckling devices and ٨٣,٠٥٪ showed that the presence of anti-suckling devices without milking as the main causes of mastitis. This finding was confirms by Abdurahman *et al*. (١٩٩٥) and Obied *et al*. (١٩٩٦); they considered this factor the major predisposing factor for camel mastitis.

In the present study, ٥٥,٠٨٪ of owners showed that the constrain of camel rearing were feed shortage and water scarcity and ٣١,٣٦٪ of them said that diseases were the main problem. Also, ٥٧,٦٢٪ used antibiotics for mastitis treatment but ٨,٤٧٪ did not practice the treatment of mastitis. This findings confirm with the findings of Abdel Gadir (٢٠٠١) who reported that ٥٪ of owners showed that the major

constrain of camel rearing were feed shortage, water scarcity and ٠٪ for diseases. However, ٥٪ of them used antibiotics for mastitis treatment but ٦٥٪ did not practice the treatment for mastitis.

This study revealed that ٨٥,٥٩٪ of the studied herds did not receive any veterinary and extension services. The results of our study revealed that the prevalence of clinical mastitis was ٠,٩٣٪ and this was in agreement with Molla (٢٠٠١), but these findings were lower than those reported by Obeid (١٩٨٣), Salwa (١٩٩٥), Bakhiet *et al*. (١٩٩٢) and Amel(٢٠٠٣) they reported values of ١٩,٥٪, ١٣,٠٩٪, ٤٥٪ and ٦,١٥٪, respectively. The disagreement of our findings with other researchers is due to the fact that other researchers collected milk samples only from she-camel suspected with mastitis.

In the present study the prevalence of sub-clinical mastitis represented a level of ٤٥,٣٧٪ and these results were in agreement with that mentioned by Obeid (١٩٨٣) who reported that an incidence rate on basis on C. M. T. was ٤٧٪. Nevertheless, these prevalence were higher than that reported by Suheir (٢٠٠٤), Amel (٢٠٠٣), Abdel Gadir (٢٠٠١), Almaw and Molla (٢٠٠١), Sana (٢٠٠٠) and Mustafa *et al*. (١٩٨٧) they reported that level of occurrence as ٤٢,٢٪, ٤٠٪, ٤١,٦٪, ٤٢,١٪, ٤١,٨٪ and ٤١,٨٪, respectively. The high prevalence of camel mastitis in this study could be due to use of anti-suckling devices which was a major predisposing factor for udder infection. So that our questionnaire survey revealed that ٩٦,٦١٪ of camel

milkers use such device. Or might be due to heavy infestation of the udder with ticks that were recorded in our sampling questionnaire a level reaching ١٠٠٪ . This factor was confirmed by Bitter (١٩٨٨), who found that tick infestation was highest in Bisharian breed of camels in the rainy season. Also, could be due to unhygienic management during milking, so that our general questionnaire survey recorded ٩٧,٤٦٪ of the camel milkers, who not clean the udder before milking and ٩٣,٣٧٪ did not wash their hands before milking. This finding was confirmed by Abdel Gadir (٢٠٠١) who found that ١٠٠٪ did not clean the udder before milking and ٥٥٪ did not wash their hands before milking the she-camels.

Our study explained that ٤٥,٨٣٪ showed the positive growth of bacteria and these findings were in agreement with the results reported by Bakhiet *et al.* (١٩٩٢), Abdurahman *et al.* (١٩٩٥) and Suheir (٢٠٠٣). They found ٤٥٪ , ٤٥٪ and ٥٤,٣٨٪ respectively. On the other hand, these results were lower than those recorded by Salwa (١٩٩٥). And Guliye *et al.* (٢٠٠٢), were ١٠٠٪ and ٨١,٤٪ rate of isolation respectively. This high result was due to collection of samples only from mastitic cases. Furthermore, this result showed that ٥٤,١٧٪ of quarter milk samples were negative for bacterial growth and this is in agreement with Amel (٢٠٠٣) and Abdurahman *et al*. (١٩٩٥), they reported that ٣٢,٦٩٪ and ٤٨,٣٪ respectively as negative for bacterial growth.

In this study the results of the W. S. T. were ١٦,٥٪ positive and this was lower than that obtained by C. M. T. according to Suheir (٢٠٠٤), Amel (٢٠٠٣) and Obeid (١٩٨٣). They reported higher positive values and they were ٣٦,٨٧٪, ٣٥,٣٨٪ and ٤٧,٣٪, respectively; and this is agreement could be due to the efficiency of the W.S.T. which is less than C. M. T. and this is according to American Public Health Association (١٩٦٠). The same source reported that variations of the W. S. T. include the C. M. T. in which a solution of the surface active agent substituted for the alkali of the original method.

Our study revealed that strong correlation between S. C. C. and bacteriological examinations. This is in agreement with Abdel Gadir (٢٠٠١) who found slight agreement between S. C. C. and bacteriological examinations results. However, Radostits *et al*. (٢٠٠٠) indicated S. C. C. rises during the first two days after another infection and returns to normal over the next ten days. Also, Salah (٢٠٠٠) confirmed that high S. C. C. in non-infected quarters could be due to the lesions on the teats and the surface of udder of the examined she-camel. Therefore, our sampling questionnaire showed that ٥٦,٦٧٪ of samples had teat lesions. Furthermore, this study showed that strong association between W. S. T. and bacteriological findings and this in agreement with Saad and Thabet (١٩٩٣). They reported strong correlation between W. S. T. and bacteriological results for mastitic camel's milk samples.

On the other hand, our study revealed that no relationship between W. S. T. and S. C. C. but when we compared these results to the results of Abdel Gadir (٢٠٠١) who found relationship between the results of C. M. T. and S. C. C., such contrast could be due to less sensitivity of the W. S. T., also, according to American Public Health Association (١٩٦٠) which reported that variations of the W. S. T. include the C. M. T. in which a solution of the surface active agent substituted for the alkali of the original method.

Staphylococcus aureus and coagulase-negative staphylococci were the most bacteria isolated from quarter milk sample suffering from sub-clinical mastitis. In the study staphylococci represented ٨٠,٣٪ of the total isolates. These results disagreed with the results reported by Amel (٢٠٠٣) and suheir (٢٠٠٤); they reported ٥٨,٨٤٪ and ٤١,٤١٪, respectively, and this high result could be due to absence of hygienic measurement during milking.

In the present study *Staphylococcus aureus* represented ٢٢,٧٥٪ of the total isolates and this was similar to those reported by Suheir (٢٠٠٤), Abdel Gadir (٢٠٠١) and Barbour *et al*. (١٩٨٥); which were represented as ٢٠,٢٪, ٢٤,٧٪ and ٢٤,٣٣٪, respectively. On the other hand, Obeid (١٩٨٣) and Salwa (١٩٩٥) reported that, the prevalence of this organism was ١٦,٩٪ and ٢٠٪, respectively. Moreover, *Staphylococcus aureus* was the main causative agent of sub-clinical mastitis and

this in agreement with the findings of Suheir (٢٠٠٤), Abdurahman *et al*. (١٩٩٥) and Amel (٢٠٠٣).

Our results revealed that *Streptococcus dysagalactiae* represented ١,٥٢٪ of the total isolates and these results was lower that those reported by Suheir (٢٠٠٤) and Amel (٢٠٠٣) which were represented as ١٧,٣٩٪ and ١٣,٦٪, respectively and this contrast could be due to the system of collection of samples which depends on random sampling technique.

This study resulted that *Corynebacterium bovis* was isolated from quarter milk samples and represented as ٣,٠٣٪ and this finding is similar to that of Abdel Gadir (٢٠٠١), Almaw and Molla (٢٠٠٠) and Abdurahman *et al*. (١٩٩٥); they isolated this bacteria from mastitic she-camel's milk. However, Corynebacterium species were isolated from mastitic milk by Suheir (٢٠٠٤), Salwa (١٩٩٥) and Amel (٢٠٠٣); the results were represented as ٧,٠٧٪, ٢,٧٪ and ٣,٣٣٪, respectively.

Bacillus cereus was isolated in this study and represented as ٩,٠٩٪ of the total isolates. This bacterium was also reported by Ramadan *et al*. (١٩٨٧), Hafiz *et al*. (١٩٨٧) and Salwa (١٩٩٥) as the causative agent of all types of she-camel mastitis.
In this study Gram-negative bacteria *Pasteurella haemolytica* was isolated from cases of clinical and sub-clinical mastitis.The results were in agreement with those of Bekele and Molla (٢٠٠١) who isolated *Pasteurella haemolytica* from sub-

clinical mastitis cases. Nevertheless, Radostits *et al*. (٢٠٠٠) considered this bacteria as uncommon pathogens that cause sporadic and sever mastitis.

The sensitivity test showed that *Staphylococcus aureus* is highly sensitive to Ciprofloxacin, Oflaxacin, Cephalexine, Tetracycline, Gentamycin, Ampicillin and moderately to Cefotaxime but resistant to Cloxacillin and Linocomycin. This finding agreed with finding of MC-Donald and Anderson (١٩٨١) who reported the sensitivity of *S. aureus* to Cephalexine, Cloxacillin, Gentamycin and Ampicillin. Also these finding are in agreement with Obeid (١٩٨٣), Abdel Gadir (٢٠٠١) Amel (٢٠٠٣) and Suheir (٢٠٠٤). They reported that Tetracycline, Gentamycin and Ampicillin were effective drugs against Camel mastitis. This finding disagreed with Buxton and Fraiser (١٩٧٧) who reported the resistance of *S. aureus* to Tetracycline, also with MC-Donald and Anderson for Cloxacillin which was sensitive and this disagreement could be due to treatment of she-Camels by Cloxacillin more frequent and lessly to Tetracycline.

In conclusion, infections with mastitis were prevalent in lactating she-camels in North Kordofan State. However, mastitis had economic importance which resulted in reduced milk production. Furthermore, camel's milk is considered as the main source of food and water for the nomadic tribes in the desert and semi desert areas. Therefore, great funds should be directed towards this area for future investigations.

Based on that, it is strongly recommended that much work should be done in clinical mastitis of lactating she-camels. The extension veterinary services must be

carried out for camel owners in order to be a ware of hygienic measurement before milking of lactating she-camel. Control of tick infestation should be carried out to reduce heavy tick infestation. Anti-suckling devices should be substituted with other means to avoid teats injuries.

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